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PubMed Central**Improved method for rapid purification of protein kinase from streptomycetes.****Janecek J, Dobrova Z, Moravec V, Naprstek J.**

Institute of Microbiology, Czech Academy of Sciences, Prague, Czech Republic.

Protein kinase from *Streptomyces lincolnensis* was purified nearly to homogeneity using a high performance liquid chromatography (HPLC) and a Pharmacia FPLC system. The procedure used employed column chromatography on DE-53, followed by FPLC affinity chromatography with serine- or threonine-Sepharose (prepared as described in this paper) and gel filtration using a Superose 12 or TSK G3000SW column. Starting with 3.5 g of mycelial proteins, approximately 1 mg of pure enzyme was obtained. The procedure is simple and highly reproducible. The protein kinase thus obtained was nearly pure by silver staining after sodium dodecyl sulfate-polyacrylamide gel electrophoresis. The purified protein kinase phosphorylated substrate proteins at the seryl residues.

PMID: 8926341 [PubMed - indexed for MEDLINE]

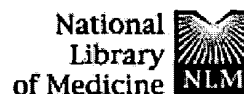
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PubMed Central**A high-yield method for the isolation of hydrophobic proteins and peptides from polyacrylamide gels for protein sequencing.****Feick RG, Shiozawa JA.**

Max-Planck Institut fur Biochemie, Martinsried, Bundesrepublik Deutschland.

A methodological approach is described which allows the isolation of hydrophobic and hydrophilic proteins and peptides in high yield. The technique consists of (1) preparative sodium dodecyl sulfate-polyacrylamide gel electrophoresis, (2) protein elution from polyacrylamide gels with an organic solvent mixture composed of formic acid/acetonitrile/isopropanol/H₂O (50/25/15/10, v/v/v/v), and (3) purification of eluted proteins by size exclusion chromatography on a Superose 12 column using this organic solvent mixture as eluant. The efficiency of this technique was tested with radioactively labeled polypeptides. These proteins were reaction center from *Chloroflexus aurantiacus*, bacteriorhodopsin, halorhodopsin from *Halobacterium halobium*, bovine serum albumin, ovalbumin, alpha-chymotrypsinogen A, and cytochrome c. The elution recoveries from polyacrylamide gels were 77-95%; the final yield after chromatographic purification was still 67-76% (with one exception). Subsequent amino acid sequencing was possible without further sample treatment. The sensitivity of the method described was found to be at least 20-30 micrograms protein.

PMID: 2382824 [PubMed - indexed for MEDLINE]

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